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TECHNICAL REPORT
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DEGRADATION OF PINK WATER COMPOUNDS IN SOIL - TNT, RDX, HMX

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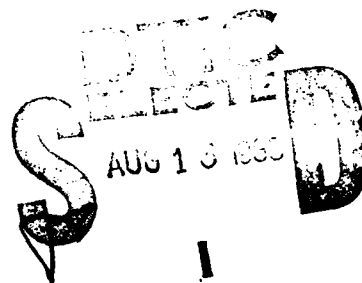
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20. ABSTRACT (Continue on reverse side if necessary and identify by block number) The degradation of pink water compounds in soil was studied. A simulated pink water containing TNT, RDX, HMX and 2,4-DNT was continuously applied to a series of soil columns at different flow rates, with and without carbon supplementation. Concentrations of TNT, RDX, HMX, total organic carbon, nitrates, nitrites, ammonia, pH, oxidation-reduction potential, and biotransformation products were monitored on a weekly basis. The results indicate that land treatment or land farming of pink water should not be considered as a treatment option for pink water. Hazardous biotransformation intermediates and		

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unchanged concentrations of some of the pink water compounds would contaminate groundwaters and soils



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PREFACE

In a recent report published by the US Army Armament Research and Development Center, "In-Plant Reuse of Pollution Abated Waters (Phase 1. Milan AAP), March 1983," land application of pink water was recommended as a possible final treatment step for contaminated wastewaters. Since this could be a low cost and simple method to alleviate pollution hazards associated with these wastewaters, the US Army Toxic and Hazardous Materials Agency requested Natick R&D Center to undertake a study of the potential for land treatment of pink water in order to address this recommendation.

This work was performed for the US Army Toxic and Hazardous Materials Agency under project number 1L161102AH68, 691000.H6800. This report covers the limited studies performed on this project over approximately five months with FY84 money. The project was terminated prematurely due to administrative decisions at Natick and therefore could not be carried through to the appropriate end-point.

We wish to thank Dr. Joseph Akkara, Steven LaRosa and Silvino Sousa, all of the Science and Advanced Technology Laboratory at the US Army Natick Research and Development Center for their technical assistance.

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DEGRADATION OF PINK WATER COMPOUNDS IN SOIL

INTRODUCTION

The environmental fate of pink water compounds has been studied primarily in water systems. Pink water is a generic term used to refer to the coloration of process waters which may contain some of the following compounds, 2,4,6-trinitrotoluene (TNT), 2,4-dinitrotoluene (2,4-DNT), hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX), and octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX). Detailed research on the degradation of TNT, 2,4-DNT, RDX and HMX in aqueous and sewage systems has been published.¹⁻⁹ In addition, the fate of TNT in composting systems was reported. However, little information is available on the degradation of these pink water components in soils, although a number of reports have documented and characterized extents of soil contamination.

An extensive literature search and review was undertaken in order to determine the extent of available data. This review included manual and computer data base searches through Natick R&D Center resources, and data bank searches through the Hazardous Materials Technical Center, Rockville, MD. The data banks searched included DIALOG, Toxline, NTIS, CA Search, Arthur Little Online, Agricola, Conference Papers Index, Enviroline, Environmental Bibliography, Pollution Abstracts, Federal Research in Progress, DROLS, EI Engineering Meetings, Compendex and Claims/US Patents. The key words searched included the following: pink water, RDX, cyclonite, TNT, waste, stability, biodegradation, disposal, fate, treatment, persistence, soil, sediment, land.

No in-depth studies were uncovered that detailed the fate of these compounds in soils, other than what was already referenced above.

In a recent Armaments R&D Center report,¹⁰ In-Plant Reuse of Pollution Abated Waters: Phase I, Milan Army Ammunition Plants (AAP), March 1983, land application of pink water was recommended as a possible treatment option. This would be a low-cost solution to this pollution problem and was recommended primarily as a final polish after pretreatment at the plant.

There is a need to fully understand the fate of pink water compounds in soils in order to properly address the above treatment option. Since the available literature is inadequate in this regard, it was the objective of this study to evaluate the degradation of pink water compounds in soils.

MATERIALS AND METHODS

Chemicals

Commercially available TNT (Eastman Kodak, Rochester, NY) was recrystallized. Dr. John Cornell, Natick R&D Center, synthesized samples of 2-amino-4,6-dinitrotoluene, 4-amino-2,6-dinitrotoluene, 2,4-diamino-6-nitrotoluene, 2,6-diamino-4-nitrotoluene, 2,2',6,6'-tetranitro-4,4'-azoxytoluene, 4,4',6,6'-tetranitro-2,2'-azoxytoluene, hexahydro-1,3,5-

trinitroso-1,3,5-triazine, and octahydro-1-nitroso-3,5,7-trinitro-1,3,5,7-tetrazocine. Hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX) and octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX) were obtained from Radford AAP Radford, VA and recrystallized 2,4-dinitrotoluene (2,4-DNT) and 1,3,5-Trinitrobenzene (TNB) were purchased from Eastman Chemical, Rochester, NY. Fig. 1 illustrates the structures for these compounds.

Soil Columns

Six glass columns (40 cm tall x 7 cm diameter) were each filled with 1000 mg dry weight of soil (garden soil passed through a 2-mm sieve, 6.5% organic matter by ignition). The soil was retained in the columns with polyethylene screening (202 μ m pore size). The pink water solutions were continuously pumped (Rainin Rabbit peristaltic pump) through silicone tubing onto the tops of the columns. The feed solutions leached through the soil via gravity. See Table 1 for soil column conditions and pink water components.

TABLE 1. Experimental conditions for the soil columns

Column Number	Microorganisms	Flow Rate	Medium	Carbon, Supplement
1	Active	Fast ^b	Tap water	None
2	Active	Fast	Pink water ^d	None
3	Active	Slow ^c	Pink water	None
4	Sterile ^a	Fast	Pink water	None
5	Active	Fast	Pink water	Glucose ^e
6	Active	Slow	Pink water	Glucose

^a0.5% mercuric chloride

^b100 mL per day

^c40 mL per day

^dSimulated pink water consisting of tap water with the following per liter: TNT, 70 mg; RDX, 30 mg; HMX, 4 mg; 2,4-DNT, 0.01 mg.

^e2.0 g per liter glucose

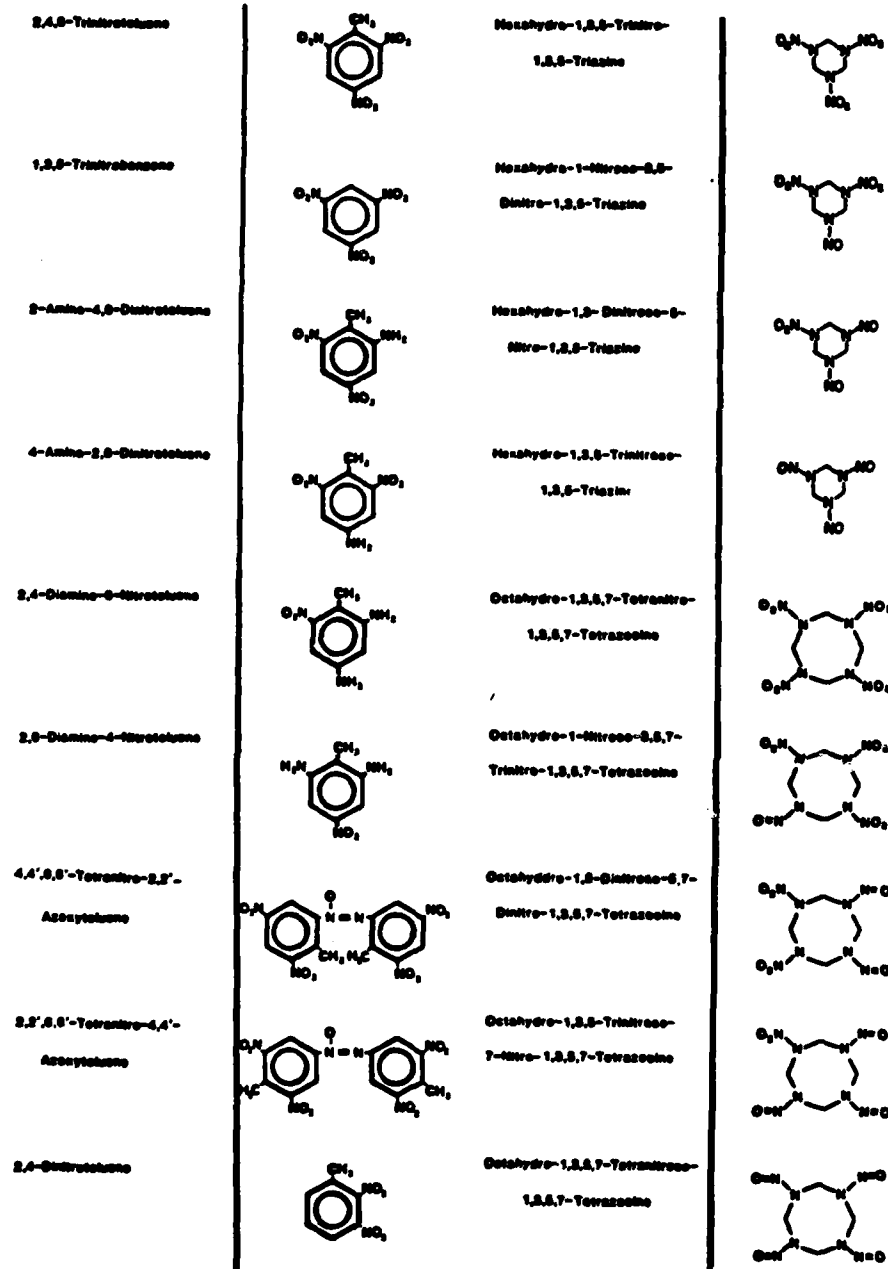


Figure 1. Structures of pink water compounds and possible intermediates.

All active columns were inoculated. The organisms were from activated sludge from the Marlborough Easterly sewage treatment plant (Marlborough, MA), anaerobic sludge digest from the Nut Island sewage treatment plant (Boston, MA), and garden soil. The combined samples were diluted with 0.85% KCl solution, filtered, and the filtrate was used as inoculum.

To make up the simulated pink water feed and minimize decomposition of some of the compounds when autoclaved, the following procedure was used. TNT and RDX were dissolved in a minimum volume of acetone in a 6-liter Erlenmeyer flask. The acetone was volatilized off with a stream of nitrogen to leave a thin film of the compounds over the surface of the glass. Sterilized and boiling tap water, 5 liters, and 2,4-DNT were added aseptically. The mixture was stirred and heated below boiling until all compounds were dissolved. This solution was then added to an autoclaved carboy containing 5 liters of tap water. When glucose was required in the feed, 1 liter of a sterile stock solution containing 2 g of glucose per liter was added.

Sampling and Analysis

Weekly samples [influent and effluent (leachate)] from the soil columns were evaluated for oxidation-reduction potential, pH, nitrites, nitrates ammonia, total organic carbon (TOC), and pink water parent compounds and intermediates.

Oxidation-reduction potential was determined with a redox platinum electrode (Orion) and reported in relative millivolts. Hydrogen ion concentration was determined with a calomel pH reference electrode and pH sample electrode (Corning). Both pH and redox were monitored on a Corning Model 130 pH meter. Low concentrations [<1 ppm (mg/L)] of nitrate were determined by a standard colorimetric method using a Lambda 3 spectrophotometer (Perkin Elmer, Norwalk, CT). Higher concentration of nitrites and nitrates were determined by high performance liquid chromatography (HPLC). Concentrations of pink water components and intermediates were also determined by HPLC.

Ammonia determinations could not be made directly on samples using a specific ion electrode due to interferences from the pink water components upon producing the alkaline pH required for the analysis. Therefore, samples for ammonia determinations were distilled to remove the ammonia from these interferences and the distillate was analyzed with an ammonia specific ion electrode (Orion).

TOC was determined with a Beckman 915B Tocamaster with Matheson Ultra Zero Air as carrier gas, flowing at 300 mL per minute. Samples, 20 μ L, were delivered with a Hamilton CR-200 200 μ L constant rate syringe.

High Performance Liquid Chromatography

A methanol/water gradient run was used to separate and quantify the various parent compounds and intermediates. The system consisted of a M721 system controller, M730 data module, M440 absorption detection set at 229 nm,

two 6000A solvent delivery systems, and a M710B sampling processor (Waters Associates, Milford, MA). The solvent flowed at 2.5 mL per min. through a C8 cartridge (8 mm diameter x 10 cm long) set in a radial compression separation system 2 module. The gradient run was initiated at 15% methanol in water for 8 minutes, programmed up to 40% methanol in water by 30 minutes, maintained at 40% until 35 minutes, and then reduced to 15% by 42 minutes. Sample injection volumes were 20 μ L and 200 μ L. Detection limits on the gradient analysis were 20 mg for TNT, RDX and HMX using a 200- μ L injection. In addition, isocratic runs with 55% and 75% methanol in water were run to further separate compounds closely eluting.

Figure 2 illustrates the separation of the TNT, RDX, HMX, and 2,4-DNT (2,4-DNT is not detectable at the 0.01 mg/L concentration under the conditions of gradient run although its retention time is indicated). Fig. 3 illustrates the same gradient analysis with separation of some of the possible intermediates. The isocratic runs were used to separate 2-amino-4,6-dinitrotoluene from 4-amino-2,6-dinitrotoluene and the azoxy intermediates. As previously reported,¹¹ baseline separation of the 2-amino-4,6-dinitrotoluene from 4-amino-2,6-dinitrotoluene could not be achieved. The azoxy compounds do not elute under the conditions of the programmed gradient analysis.

Nitrate and nitrite were separated and quantified by HPLC. The solvent system consisted of 2.5 mM phosphate buffer flowing at 3 mL per minute through an anion exchange column, SAX, 10 μ m packing (8 mm diameter x 10 cm long) radial compression cartridge (Water Associates). The anions were monitored at 229 nm. The retention times were approximately 6 minutes and 5 minutes for nitrate and nitrite, respectively. Injection volumes were 25 μ L or 200 μ L.

Bioassays

Bioassay work on the simulated pink water, influent medium, soils, and leachates from the soil columns, was performed by US Army Environmental Hygiene Agency (USAEHA) under the direction of Henry Eichorn. Daphnia and algal tests were run according to standard procedures.^{12,13} However, due to repeated delays at USAEHA, no data was available at the time of publication of report. Data will be made available at a later date through the contracting office, the US Army Toxic and Hazardous Materials Agency (USATHAMA).

RESULTS

The soil columns were run continuously for 110 days. In all subsequent figures the results are presented for all columns listed in Table 1 with the exception of the control column (#1 - tap water feed, no pink water). No significant interferences appeared due to contamination of the tap water or in leachates from the soil used in this column.

Figure 4 presents the recovery data for TNT. Percent recovery of TNT indicates that almost all of the TNT fed into the columns had either remained in the soil or been biotransformed. Only in column #2 (fast flow, no carbon supplement) does there appear to be significant breakthrough of TNT during the time frame of these experiments.

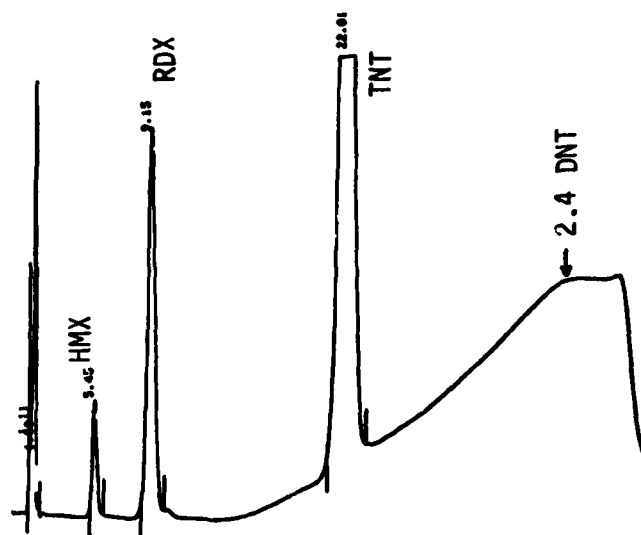


Figure 2. Gradient analysis of simulated pink water containing 70mg/L TNT, 30mg/L RDX, 4 mg/L HMX, and 0.01mg/L DNT.

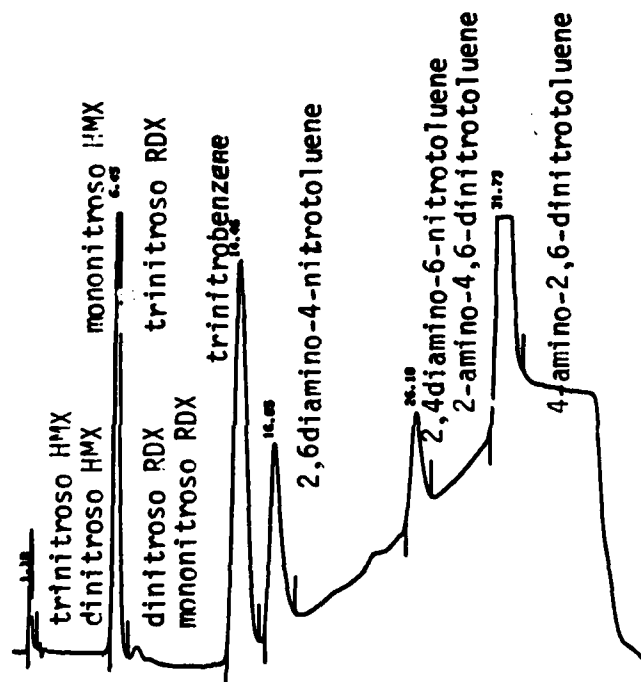


Figure 3. Gradient analysis of some of the potential intermediates.

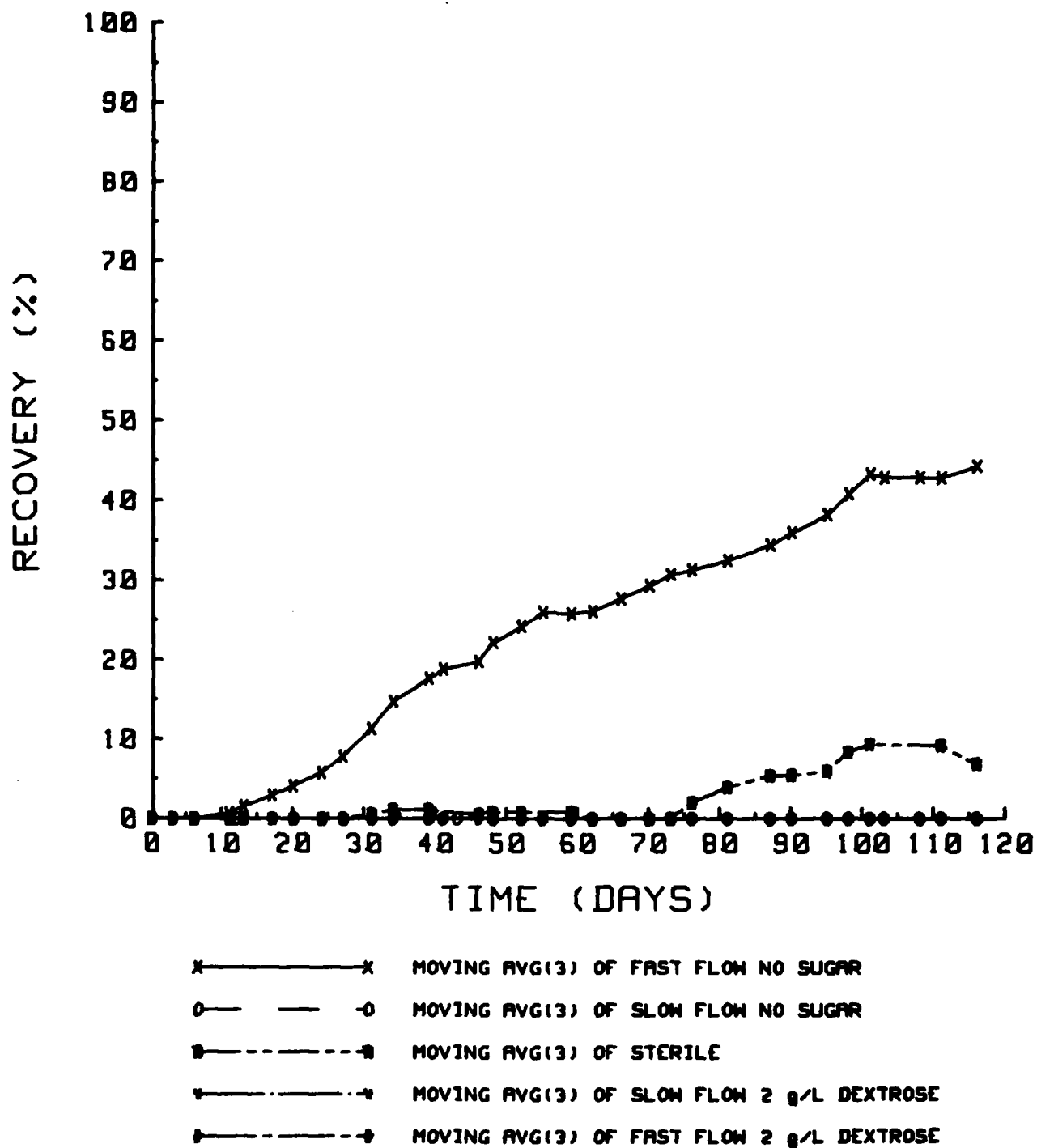


Figure 4. Recovery of TNT from soil columns.

In both column #2 (fast flow without carbon supplement) and column #5 (fast flow with carbon supplement) the microbially produced intermediate, 4-amino-2,6-dinitrotoluene, was identified. In column #5, 2-amino-4,6-dinitrotoluene was also identified. These results would indicate that microbial activity in the columns is at least in part responsible for the low recovery of TNT in the leachates. These results also indicate that supplemental carbon is not a requirement for activity in the biotransformation of TNT.

Fig. 5 presents recovery data for RDX from the simulated pink water. Only in column #6 (slow flow with carbon supplement) does there appear to be a significant decrease in RDX recovery, indicating microbial activity. RDX is not readily bound or retained in the soil as evidenced by its early breakthrough in column leachates. It also appears that the recovery pattern for column #5 (fast flow with carbon supplement) indicates a similar trend as that observed for column #6 (slow flow with carbon supplement).

In column #5 (fast flow with carbon supplement) both mononitroso- and dinitroso-RDX were identified in leachates. In leachates from columns #3 (slow flow without carbon supplement) and #6 (slow flow with carbon supplement) mononitroso-RDX was identified.

Fig. 6 illustrates the recovery data for HMX and its rapid breakthrough from the soil columns. Within the time frame of the study there appears to be little evidence for degradation of HMX under any of the column conditions studied. No intermediates from HMX were detected in column leachates.

Note that 2,4-DNT was not detectable at the low levels used in the simulated pink water; therefore, no recovery data is presented. However, 2,4-DNT was still included in the simulated waste stream in order to assess its possible effects during bioassay studies on the exposed soils.

Fig. 7 illustrates nitrate and nitrite data from the active control, column #1 (fast flow, no carbon supplement). No appreciable levels of these anions were present in the tap water or in leachates from the soil used in the study.

Results of nitrate evaluations from the remaining columns are illustrated in Fig. 8. Influent (left graph) concentrations are all below 10 ppm, while results of analysis of leachates (right graph), indicate levels up to around 40 ppm in columns with carbon supplementation (#5 and #6) and in column #3 (slow flow, no carbon supplement). Levels of nitrate in leachates from column #1 leveled off at around 20 ppm, while the leachates from the sterile control (column #4) contained approximately the same levels of nitrate as in the influent. These results indicate that columns with the greatest microbial activity are also the columns with the highest nitrate levels appearing in leachates.

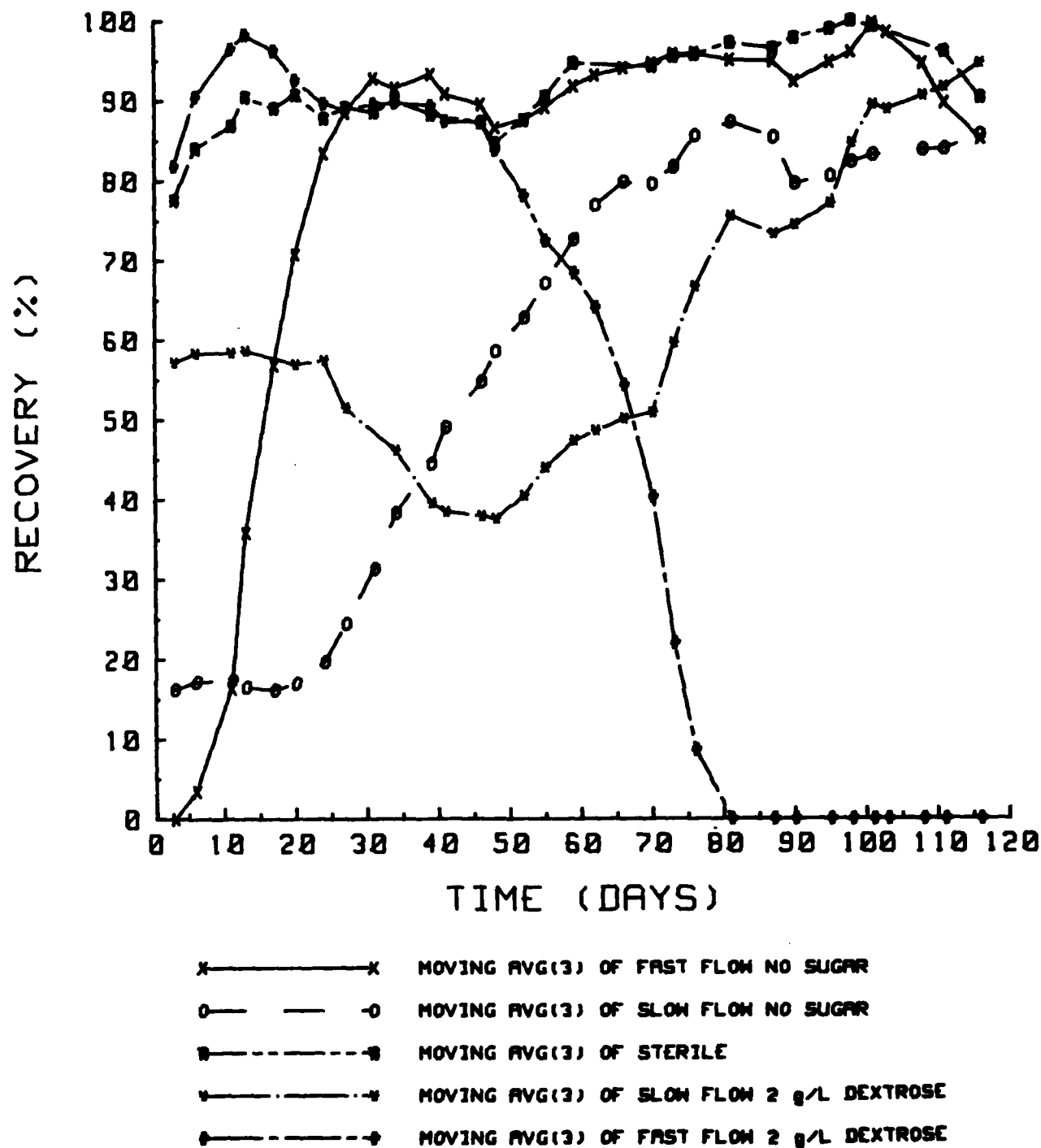


Figure 5. Recovery of RDX from soil columns.

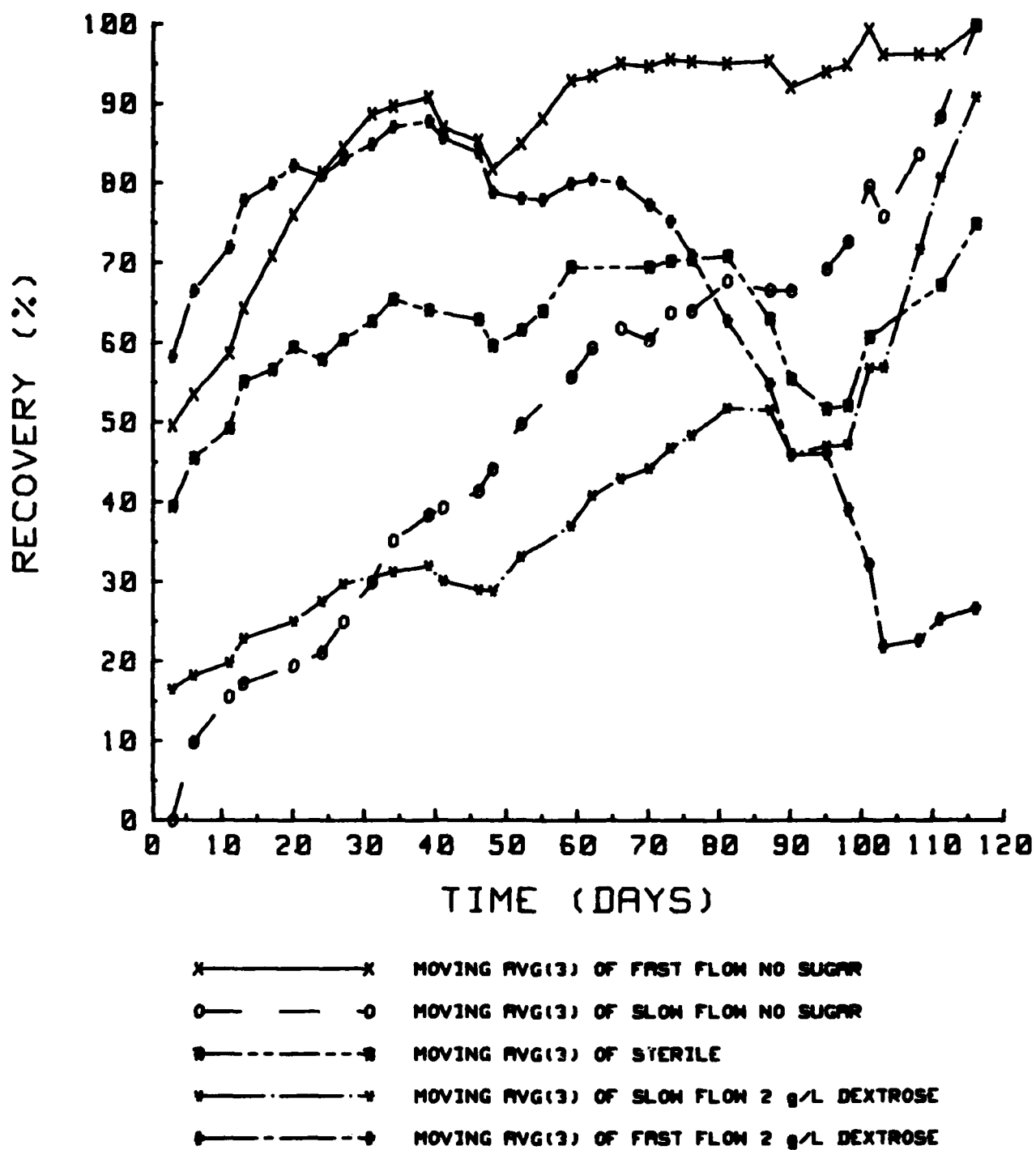


Figure 6. Recovery of HMX from soil columns.

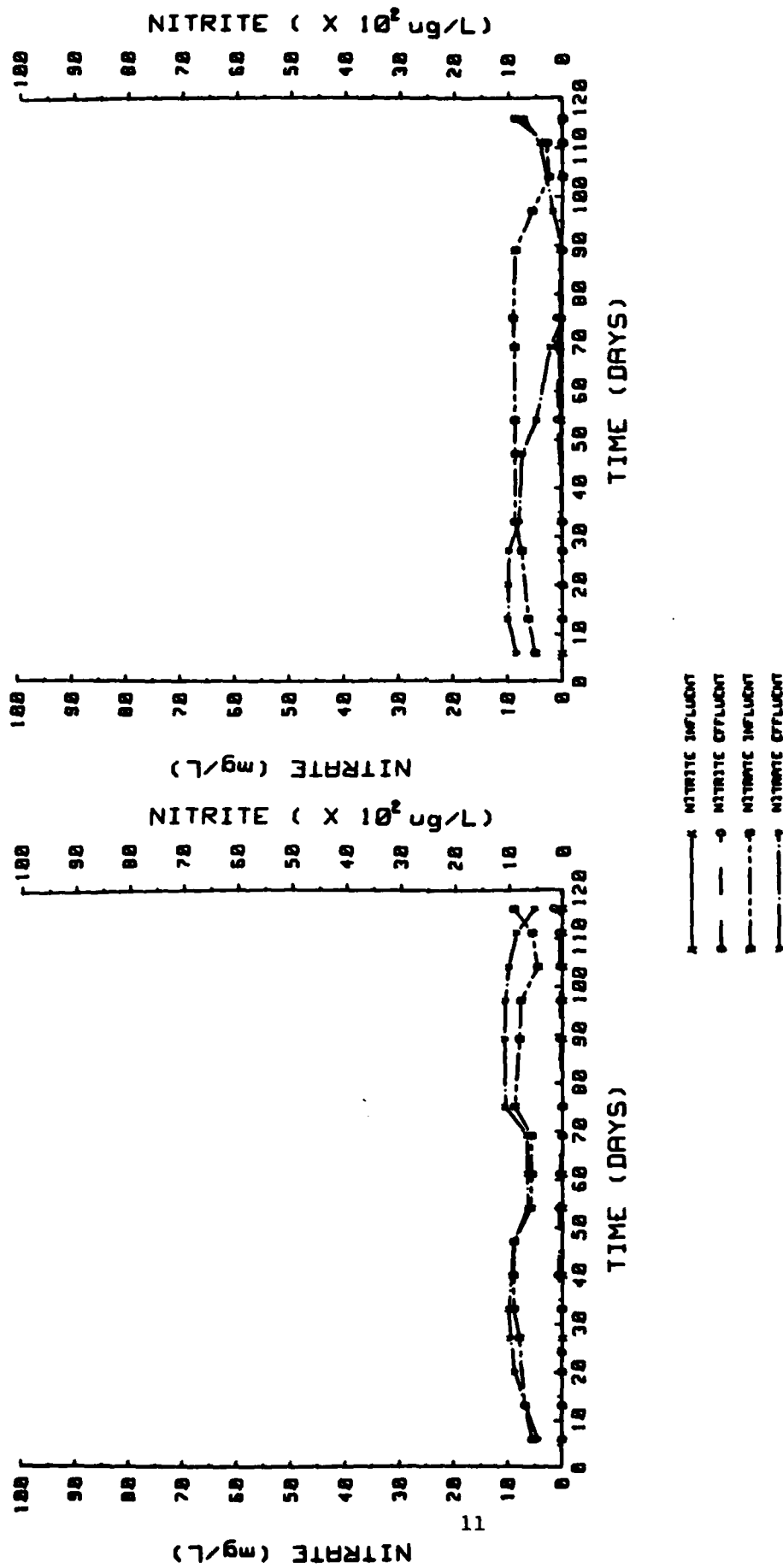


Figure 7. Nitrate and nitrite recovery data from the active control soil column without pink water (left) and the sterile control pink water column (right).

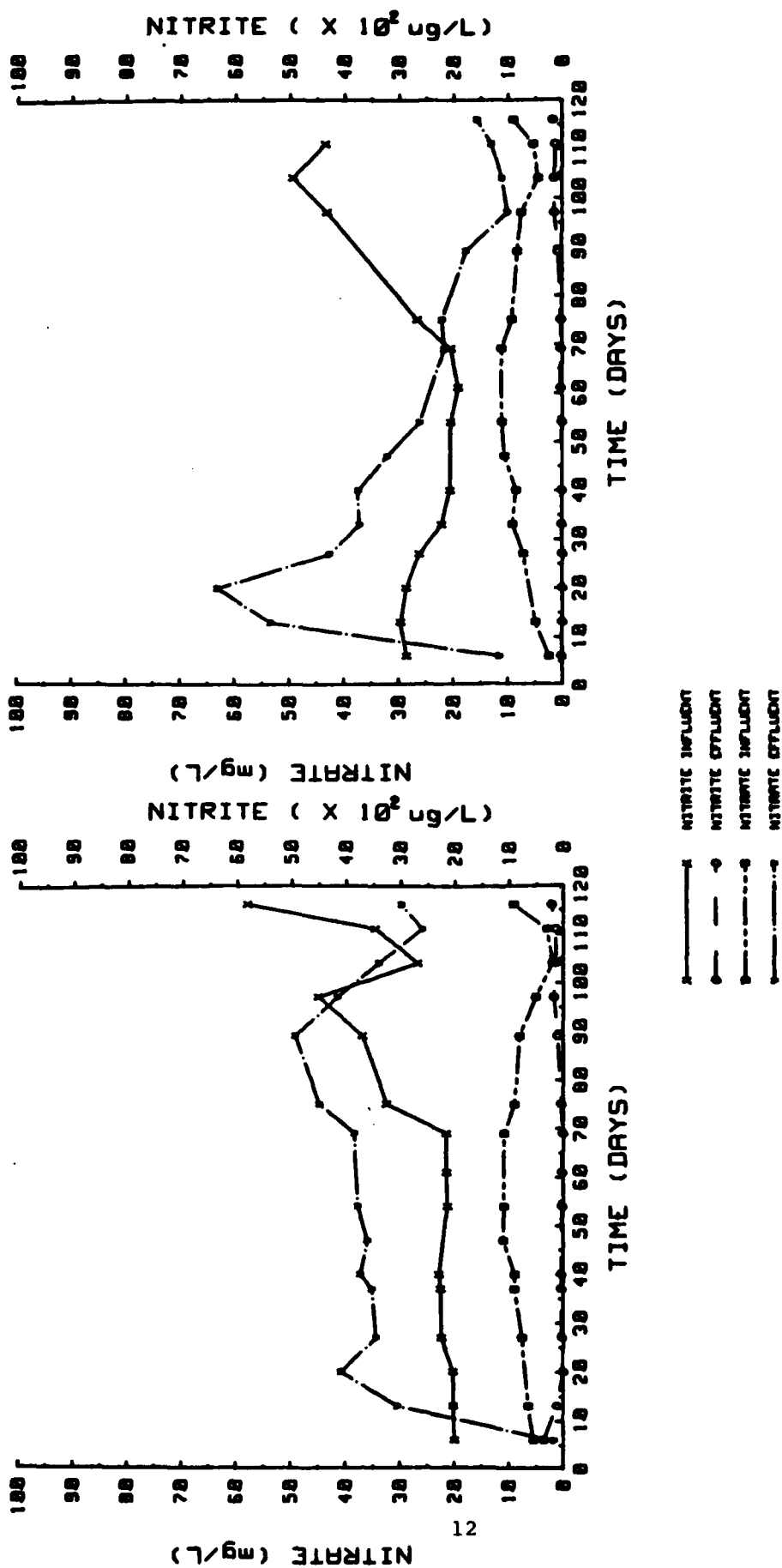


Figure 8. Nitrate and nitrite recovery data from slow flow pink water soil columns, active without carbon (left graph) and active with carbon (right graph).

Corresponding data for nitrite is presented in Fig. 9. Nitrite levels were below 3.5 ppm throughout the study for influent feed samples, while leachates contained nitrite concentrations well below these concentrations.

Ammonia concentrations, determined after distillation from interfering compounds, indicated concentrations below 0 ppm in all influent and leachate samples evaluated. Samples were examined after 700 days of equilibration of the columns.

Figure 10 illustrates the appearance of 4-amino-2,6-dinitrotoluene as a degradation product in leachates.

Figures 11 illustrates the appearance of mononitroso-RDX as a degradation product in leachates.

Results from TOC analysis are illustrated in Table 2. When glucose was not added in these systems, between 30 mg/L and 50 mg/L TOC was present in leachates from the columns. With 2 g/L glucose there was no contribution to effluent TOC in column 6, while column 5 contained somewhat elevated levels of TOC in leachates.

TABLE 2. TOC Results for the Soil Columns

Column No. ^a	Glucose (g/L)	Influent (mg/L) ^b	Effluent (mg/L) ^b	Decrease (%) ^b
1	0	23.4 ± 13.8 (12)	34.3 ± 13.7 (12)	--- ^c
2	0	37.3 ± 9.1 (12)	43.1 ± 14.8 (11)	---
3	0	41.7 ± 19.6 (11)	50.9 ± 19.0 (12)	---
4	0	42.0 ± 15.8 (12)	46.3 ± 10.7 (9)	---
5	2.0	653.2 ± 211.9 (12)	119.1 ± 137.3 (11)	83.9 ± 14.9 (11)
6	2.0	560.3 ± 176.2 (9)	49.3 ± 15.3 (12)	91.2 ± 4.7 (9)

^areference Table 1 for condition.

^baverage ± 1 standard deviation; number of samples in parentheses.

^cno decrease.

Table 3 summarizes the identification of intermediates from the different soil columns, as well as presenting total weights of the pink water compounds added to and recovered from the columns.

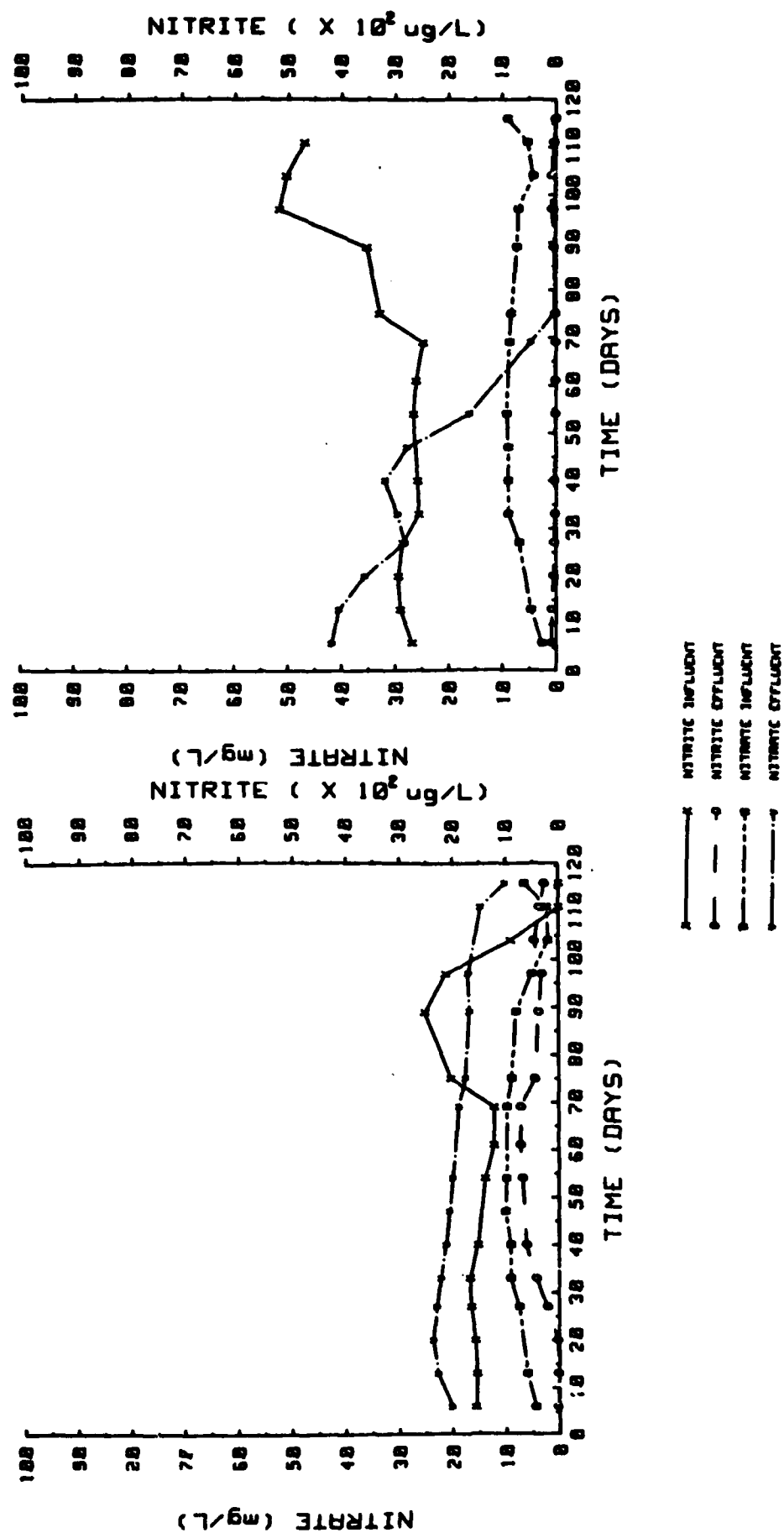


Figure 9. Nitrate and nitrite recovery data from fast flow pink water soil columns, active without carbon (left graph) and active with carbon (right graph).

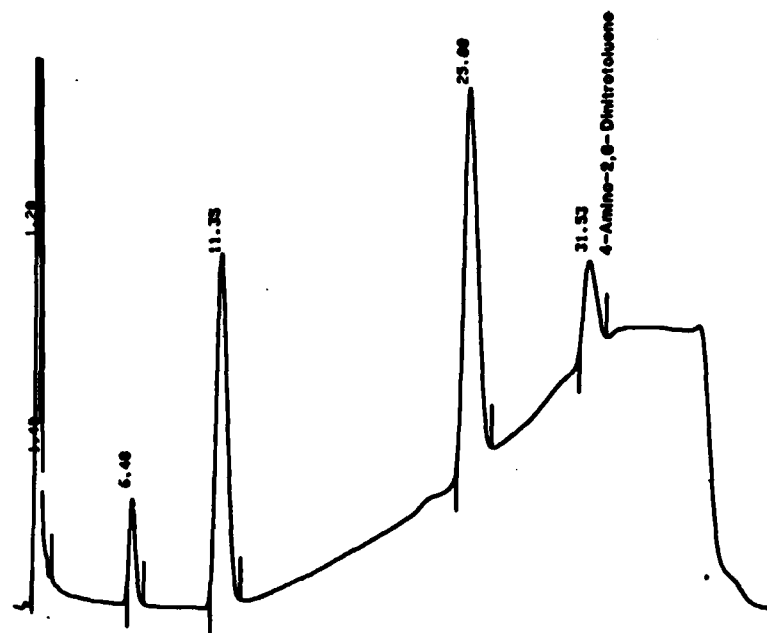


Figure 10. Gradient analysis of leachate illustrating appearance of 4-amino-2,6-dinitrotoluene.

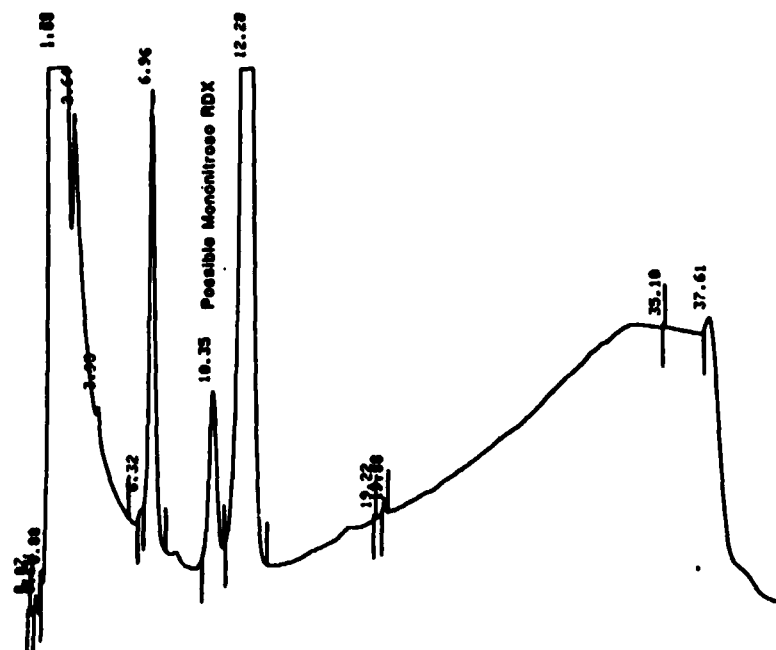


Figure 11. Gradient analysis of leachate illustrating appearance of mononitroso-RDX.

TABLE 3. Summary of Results for the Individual Soil Columns Including Intermediates Identified in Column Leachates and Total Weight of Pink Water Compounds Added to and Recovered from the Different Columns During the Study

COLUMN CONDITIONS	2,4,6-Trinitrotoluene	Hexahydro-1,3,5-Trinitro-1,3,5-Triazine	Octahydro-1,3,5,7-Tetranitro-1,3,5,7-Tetrazocine
Active Soil Pink Water Fast Flow	mg on = 1336 mg off = 320 4-amino-2,6-dinitrotoluene percent reduction = 76	mg on = 443 mg off = 372 percent reduction = 16	mg on = 52 mg off = 47 percent reduction = 10
Active Soil Pink Water Slow flow	mg on = 483 mg off = 0 percent reduction = 100	mononitroso - RDX dinitroso - RDX mg on = 138 mg off = 80 percent reduction = 42	mg on = 18 mg off = 9 percent reduction = 50
Sterile Control Pink Water Fast Flow 0.5% HgCl_2	mg on = 778 mg off = 16 percent reduction = 98	mg on = 350 mg off = 299 percent reduction = 15	mg on = 37 mg off = 36 percent reduction = 3
Active Soil Pink Water Fast Flow 2 g/L Dextrose	4-amino-2,6-dinitrotoluene mg on = 2260 mg off = 0 percent reduction = 100	mononitroso - RDX dinitroso - RDX mg on = 316 mg off = 137 percent reduction = 57	mg on = 58 mg off = 36 percent reduction = 38
Active Soil Pink Water Slow Flow 2 g/L Dextrose	mg on = 652 mg off = 0 percent reduction = 100	mononitroso - RDX dinitroso - RDX mg on = 99 mg off = 59 percent reduction = 40	mg on = 17 mg off = 7 percent reduction = 59

DISCUSSION

The results of this study, although limited to four months of research, clearly demonstrate that pink water can not be adequately treated in soil. Although TNT appears to be rapidly biotransformed or retained under the conditions of this study, microbial intermediates had begun to appear in leachates from the soil. These intermediates have been shown to be hazardous, and would therefore pose a significant threat to groundwaters and soils.^{14,15,16} The same holds true for RDX. Although some biodegradation was found under some of the study conditions, nitroso-intermediates were identified in leachates that would also pose a hazard to groundwaters and soils. HMX appeared to be unaffected by the soil conditions studied, passing essentially unchanged through the soil columns, and thus would also pose a significant threat to groundwaters.

The columns receiving carbon supplementation exhibited greater activity towards biotransforming the simulated pink water (see Table 2). The requirement for supplementation is a reflection of the cometabolism of many nitrogen-rich explosives and propellants, and was not unexpected.

During the 116-day study, the total weight of the various pink water components added to the soil, and that retained in the soil is indicated in Table 2. On a basis of 1000 g dry weight of soil, this translates into a carrying capacity of 629.8, 15.4, and 6.0 ug/g soil for this particular setup for TNT, RDX and HMX, respectively. This also translates into the fact that in order of leachability and migration in groundwaters, TNT can be expected to migrate the least and HMX the most rapidly of these three compounds.

CONCLUSIONS

During this 116-day study on the fate of simulated pink water compounds in soil, it is clear that land application is not a viable option for treatment. The simulated pink water contained TNT, 70 mg/L; RDX, 30 mg/L; HMX, 4 mg/L, and 2,4-DNT, 0.01 mg/L. The results of this study indicate that some components of the simulated waste stream pass through the soil columns unchanged during the timeframe of the study. The results also indicate that biotransformation products are formed that will present a hazard to soils and groundwaters. HMX broke through the soil columns most rapidly, followed successively by RDX and TNT. This reflects the degree of migration of these compounds in groundwaters. Supplemental carbon did stimulate the biological activity in the soil with regard to the biotransformation pink water compounds.

Since the pink water compounds were not completely degraded in the soil and hazardous intermediates appeared in leachates, land treatment of pink water should not be considered as a treatment option to dispose of pink water laden process waters. It would not be expected that longer-term studies would provide data to change the general conclusions reached during this 116-day study.

This document reports research undertaken at the US Army Natick Research and Development Command and has been assigned No. NATICK/TR-851046 in the series of reports approved for publication.

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